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INTERNATIONAL APPLICATION PUBLISHED UNDER THE PATENT COOPERATION TREATY (PCT)

(51) International Patent Classification 6 : C07D 487/04, A61K 31/40 — A1

(11) International Publication Number: WO 97/23484

(43) International Publication Date: 3 July 1997 (03.07.97)

(21) International Application Number: PCT/SE96/01679

(22) International Filing Date: 17 December 1996 (17.12.96)

(30) Priority Data:
9504618-1 22 December 1995 (22.12.95) SE

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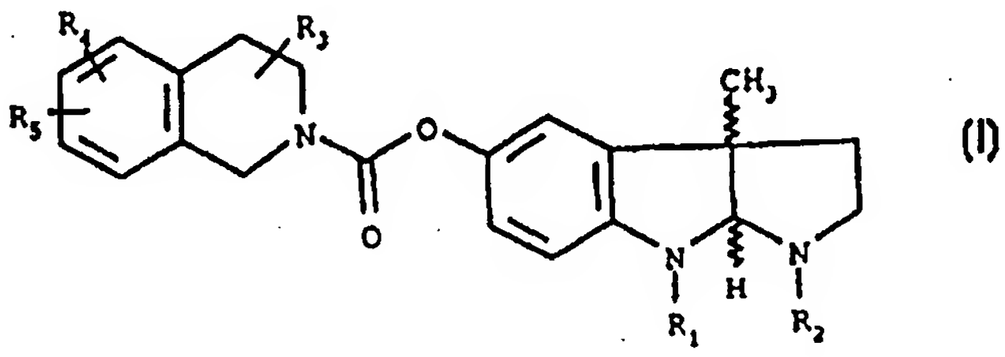
(81) Designated States: AL, AM, AT, AU, AZ, BA, BB, BG, BR, BY, CA, CH, CN, CU, CZ, DE, DK, EE, ES, FI, GB, GE, HU, IL, IS, JP, KE, KG, KP, KR, KZ, LC, LK, LR, LS, LT, LU, LV, MD, MG, MK, MN, MW, MX, NO, NZ, PL, PT, RO, RU, SD, SE, SG, SI, SK, TJ, TM, TR, TT, UA, UG, US, UZ, VN, ARIPO patent (KE, LS, MW, SD, SZ, UG), Eurasian patent (AM, AZ, BY, KG, KZ, MD, RU, TJ, TM), European patent (AT, BE, CH, DE, DK, ES, FI, FR, GB, GR, IE, IT, LU, MC, NL, PT, SE), OAPI patent (BF, BJ, CF, CG, CI, CM, GA, GN, ML, MR, NE, SN, TD, TG).

Published
With international search report.

(54) Title: NEW COMPOUNDS

(57) Abstract

The invention relates to novel tetrahydroisoquinolinyl carbamates of pyrroloindole derivatives of formula (I) having pharmacological actions and to processes for the synthesis of, and formulations containing such derivatives. It also relates to compounds which are intermediate products in the manufacture of said derivatives. These compounds which inhibit cholinesterase and are useful for enhancing memory and for treating Alzheimer's disease.



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NEW COMPOUNDS

Field of the invention

The present invention relates to novel tetrahydroisoquinolinyl carbamates of pyrroloindole derivatives having pharmacological actions and to processes for the synthesis of, and formulations containing such derivatives. It also relates to compounds which are intermediate products in the manufacture of said derivatives. More particularly, the present invention relates to compounds which inhibit cholinesterase and are useful for enhancing memory and for treating Alzheimer's disease.

Background of the invention

Acetylcholinesterase (AChE), sometimes called true or specific cholinesterase, is found in nerve cells, skeletal muscle, smooth muscle, various glands and red blood cells of mammals. AChE may be distinguished from other cholinesterases by substrate and inhibitor specificities and by distribution in the mammalian body. Its distribution in brain roughly correlates with cholinergic innervation and subfractionation shows the highest level in nerve terminals. AChE is also found in other animals, for example in the skin of the electric eel. Electric eel AChE has been used in pharmacology as a test model for human AChE.

It is generally accepted that the physiological role of AChE is the rapid hydrolysis and inactivation of acetylcholine. Inhibitors of AChE show marked cholinomimetic effects in cholinergically-innervated effector organs and have been used therapeutically in the treatment of glaucoma, myasthenia gravis and paralytic ileus. However, studies have suggested that AChE inhibitors may also be useful for alleviating memory dysfunctions characterized by a cholinergic deficit, such as Alzheimer's disease.

Physostigmine is a potent acetylcholinesterase inhibitor. However, its therapeutic utility is limited by poor stability and low oral bioavailability, short duration of action and high acute toxicity. WO90/03552 and US 5,187,165 have disclosed (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate (hereinafter referred to as C I) as having improved properties with respect to stability and

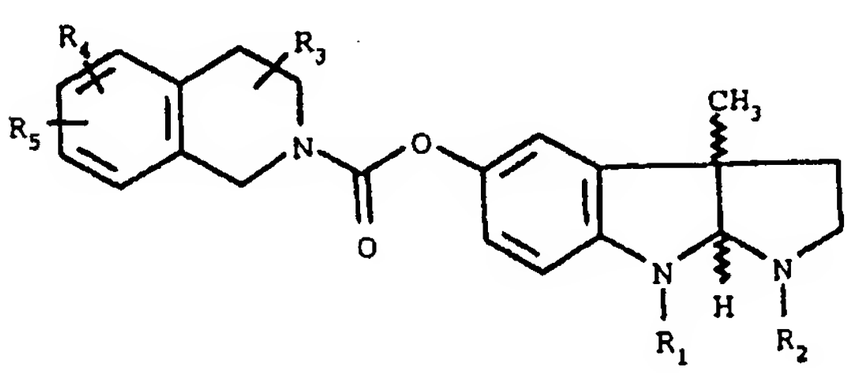
duration of action in in vitro and in vivo tests. Oral bioavailability was also improved over physostigmine in in vivo tests with rats.

Because of the high toxicity of physostigmine it is desirable to find ways of reducing such effects while still retaining the high pharmacological potency of the selected compounds. Compounds having such beneficial properties could then be formulated as drugs for enhancing acetylcholine levels in the brain. Important properties for the utility of such drugs are high degree of inhibition of AChE, good stability, long duration of action , good bioavailability, good partition across the blood brain barrier, and few side effects.

Description of the invention

The present invention provides novel compounds which are acetylcholinesterase inhibitors and which have advantageous properties with respect to stability, duration of action, oral bioavailability and/or partition across the blood brain barrier. The compounds are easily metabolized and/or excreted from in the mammalian body. The compounds are indicated for use in alleviating states of cholinergic deficit leading to memory dysfunction e.g. Alzheimer's disease. The compounds are also indicated for use in the treatment of other diseases resulting from cholinergic deficit e.g. glaucoma, myasthenia gravis and paralytic ileus.

The present invention relates to the 3a*S*- *cis* and 3a*R*- *cis* isomers or racemic mixtures or any other mixtures thereof of the compounds of formula I,



I

wherein R_1 and R_2 , which may be the same or different, are each H or CH_3, and

R_3 is H or OH,

R_4 and R_5 , which may be the same or different, are each H, OH or OCH_3,

provided that

a) at least one of R_1 and R_2 is H when R_3 , R_4 and R_5 all are H,

b) when R_1 and R_2 are both CH_3 and R_3 is OH, then R_4 and R_5 are both H,

c) when R_1 and R_2 are both CH_3 and R_3 and R_4 are both H, then R_5 is OH in position 5 of the isoquinolyl moiety of the molecule,

d) when R_1 and R_2 are both CH_3 , and R_3 is H, then R_4 and R_5 are either both OH or both OCH_3 in positions 6 and 7 of the isoquinloyl moiety of the molecule,

and pharmaceutically acceptable acid addition salts thereof.

The compounds of the present invention have chiral centres and can, therefore, exist as enantiomeric forms and as racemates. Most preferred are the 3aS-cis compounds . These forms and racemates are included in the scope of the invention.

The compounds of the present invention can be used in the form of the free base or as pharmaceutically acceptable acid addition salts. Acceptable acids for this purpose include, but are not limited to, inorganic acids e.g. hydrochloric, hydrobromic, sulfuric, nitric, phosphoric and perchloric acids as well as organic acids e.g. tartaric, citric, acetic, succinic, maleic, fumaric and oxalic acids.

As examples of pharmaceutical compositions containing the compounds of the present invention the following can be mentioned:

The compounds of the present invention may be orally administered, for example, with an inert diluent or with an edible carrier, or they may be enclosed in gelatin capsules, or they may be compressed into tablets. For the purpose of oral therapeutic administration, the active compounds of the invention may be incorporated with capsules, elixirs, suspensions, syrups, wafers, chewing gum and the like. The content of active substance of the preparations may be varied depending upon the particular form and may be between 0.5% to about 99% by weight of the dosage unit. The amount of active compound in such compositions is such that a suitable unit dosage will be obtained. Preferred compositions and preparations according to the present invention are prepared so that an oral dosage unit form contains between 0.5 mg and 5000 mg of the active compound.

Because the compounds are rapidly eliminated they can suitably be injected into the mammalian body. For the purpose of parenteral therapeutic administration, the active compounds of the invention may be incorporated into a solution or suspension. These preparations should contain between 0.5% and 30% by weight of active compound. The amount of active compound in such compositions is such that a suitable dosage will be obtained. Preferred compositions and preparations according to the present invention are prepared so that a parenteral dosage unit contains between 0.5 mg and 100 mg of active compound.

The amount of active compound to be administered will vary depending on the severity of the disease and the mode of administration, but may be in the interval of 0.5 to 5000 mg active compound per day. The AChE inhibiting activity of the compounds of the present invention was demonstrated in the following two experiments.

The compounds of the present invention can be synthesised according to the following examples:

Example 1

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate (C II).

The compound CII is synthesized by demethylation of C I via oxidation to (3a*S-cis*)-8-formyl-1,2,3,3a,8,8a-hexahydro-1,3a-dimethylpyrrolo [2,3-*b*] indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate (C IX) and hydrolysis with a strong acid acid in aqueous solution according to the following:

A mixture of C I (0.26 mmol), pyridinium dichromate (0.32 mmol), acetic anhydride (0.80 mmol) and CH_2Cl_2 (3 ml) was heated under reflux for 1.5 h. The reaction mixture was filtered through a short silica gel column with a plug of ethyl acetate above it. The residue obtained after concentration of the solvent , was purified on a silica gel column (CH_2Cl_2:CH_3OH , 50:1) giving C VI (64 mg, 164 µmol, 63%) as a clear syrup.

C IX (127 µmol) was hydrolyzed with 10% HCl (2ml) at room temperature. The solution was made basic with sat. $NaHCO_3$ and extracted with CH_2Cl_2. The organic phase was washed with brine, dried Na_2SO_4 and concentrated. The residue was purified on a silica gel column (CH_2Cl_2:CH_3OH, 10:1) giving C II (33 mg, 90 µmol, 71%).

^{13}C-NMR ($CDCl_3$) δ 154.8, 146.8, 144.5, 137.7, 134.7, 133.5, 133.1, 129.0, 128.7, 126.7, 126.5, 126.4, 120.9, 116.9, 109.4, 90.1, 54.0, 52.5, 46.3, 46.1, 42.3, 41.7, 40.7, 36.7, 29.2, 28.9, 26.9

^{1}H NMR ($CDCl_3$) δ 7.26-7.14 (m, 4H), 6.63 (d, J = 2.3 Hz, 1H), 6.77 (dd, J = 8.3, 2.3 Hz, 1H), 6.55 (d, J = 8.3 Hz, 1H), 4.81 (bs, 1H), 4,69 (bs, 1H), 4.51 (s, 1H), 3.82 (m, 2H), 2.93 (bs, 2H), 2.78 (m, 1H), 2.63 (m, 1H), 2.47 (s, 3H), 2.02 (m, 2H), 1.45 (s, 3H)

MS 363 (100, M^+), 348 (5, M^+-CH_3), 319 (25), 305 (7), 203 (25), 160 (24)

Example 2

(3a*S-cis*)-1,2,3,3a,8,8a-hexahydro-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate (C III).

This synthesis is suitable for the compounds of the present invention having a hydrogen in position 1 of the pyrroloindole moiety of the molecule. The compound is synthesized starting from (-)-N^1-benzyleseroline according to Yu, Q.-S., Atack, J.R., Rapoport, I., Brossi, A.J., J. Med. Chem. 1988, 31, 2297-2300 to give (3a*S-cis*)-1,2,3,3a,8,8a-

hexahydro-1-benzyl-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)isoquinolinecarboxylate (C VII), which is then hydrogenated by treatment with hydrogen in the presence of a palladium catalyst , resulting in C III.

The synthesis was performed as follows: To a solution of (-)-N^1-benzyleseroline (0.28 mmol) in CH_2Cl_2 (1.8 ml) was added 1,1'-carbonyldiimidazole (0.34 mmol). The mixture was stirred at room temperature for 1 h forming (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl imidazolecarboxylate after which 1,2,3,4-tetrahydroisoquinoline (0,62 mmol) was added. After 15 h the reaction mixture was concentrated to give a dark red residue , which was purified on a silica gel column (toluene:C_2H_5OAc, 2:1) to give C VII (100 mg, 0,22 mmol, 79%).

C VII (103 µmol) was dissolved in a mixture of CH_3OH:C_2H_5OAc (10:1, 4 ml), and palladium hydroxide on carbon (4 mg) was added. After the mixture was stirred for 1 h under hydrogen at room temperature, the palladium catalyst was filtered through Celite (Fuller's Earth) and the solvent was evaporated in vacuo. The residue was purified on a silica gel column (CH_2Cl_2:CH_3OH, 10:1), giving C III (29 mg, 80 µmol, 77%) as a white foam.

^{13}C-NMR ($CDCl_3$) δ 154.7, 148.3, 142.8, 136.2, 134.5, 134.3, 133.3, 132.9, 128.8, 128.5, 126.5, 126.3, 126.1, 120.5, 116.5, 105.0, 92.6, 60.3, 52.1, 46.0, 45.9, 42.2, 42.1, 41.5, 32.4, 29.0, 28.7, 26.0

MS 363 (43, M^+), 203 (100), 160 (36)

Example 3

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-4-hydroxy-2(1*H*)-isoquinolinecarboxylate (C IV).

This synthesis is suitable for compounds of the invention having a hydroxyl group in position 1, 3 or 4 of the isoquinoline moiety. The compounds are synthesized from eseroline and the corresponding 1,2,3,4-tetrahydro-hydroxyisoquinoline. Eseroline was synthesized as described by Yu, Q.-S., Schönenberger, B., Grossi, A., Heterocycles 1987, 26, 1271-1275.

1,2,3,4-tetrahydro-4-hydroxyisoquinoline was synthesized as described by Ra,, S., Saxena, A.K., Jain, P.C., Ind. J. Chem. 1978, 16B, 1019.

To a solution of eseroline (787 µmol) in CH_2Cl_2 (4 ml) was added 1,1'-carbonyldiimidazole (944 µmol). The mixture was stirred at room temperature for 1 h forming (3a*S-cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl imidazolecarboxylate after which 1,2,3,4-tetrahydro-4-hydroxyisoquinoline (1102 µmol) in CH_2Cl_2 (1 ml) was added. After 15 h the reaction mixture was concentrated to give a residue, which was purified by column chromatography (CH_2Cl_2:CH_3OH, 10:1) giving C IV (78 mg, 197 µmol, 25%) as a white foam.

^{13}C-NMR ($CDCl_3$) δ 155.4, 149.6, 143.4, 137.5, 136.6, 136.5, 133.0, 132.7, 128.3, 128.1, 127.2, 127.0, 126.8, 126.1, 126.0, 120.6, 116.3, 106.7, 97.9, 66.1, 65.9, 53.2, 52.7, 48.9, 48.4, 46.2, 45.9, 40.7, 38.3, 37.1, 27.3

^{1}H NMR ($CDCl_3$) δ 7.48-7.15 (m, 4H), 6.82 (dd, J = 8.4, 2.4 Hz, 1H), 6.77 (d, J = 2.4 Hz, 1H), 6.34 (d, J = 8.4 Hz, 1H), 5.05-4.50 (bm, 3H), 4.12 (s, 1H), 3.96 (m, 1H), 3.76 (bd, 1H), 2.92 (s, 3H), 2.64 (m, 1H), 2.61 (m, 1H), 2.53 (s, 3H), 1,94 (m, 2H), 1,42 (s, 3H)

Example 4

(3a*S-cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-5-hydroxy-2(1*H*)-isoquinolinecarboxylate (C V).

This synthesis is suitable for compounds having a hydroxyl substituent in any of positions 5, 6, 7 or 8 of the isoquinolinyl moiety of the compound. The compounds are synthesized via the corresponding methoxylated compound i.e. a compound having a methoxyl substituent in any of positions 5, 6, 7 or 8 by coupling the methoxy-1,2,3,4-tetrahydromethoxyisoquinoline with eseroline and then demethylation of the intermediate compounds with a mineral acid, or a Lewis acid, e.g. BBr_3.

Synthesis of the intermediate compound (3a*S-cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-5-methoxy-2(1*H*)-isoquinolinecarboxylate (C VIII).

To a solution of eseroline prepared from eseroline fumarate (1.176 g, 3 mmol) in CH_2Cl_2 (15 ml) is added 1,1'-carbonyldiimidazole (0.729 g, 4.5 mmol). The solution is stirred at room temperature for two hours forming (3a*S-cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl imidazolecarboxylate. 1,2,3,4-tetrahydro-5-methoxyisoquinoline (0.733 g, 4.5 mmol) is added and the solution is stirred at room temperature for 16 hours. CH_2Cl_2 (50 ml) is added. The solution is washed with water, then with brine, dried over Na_2CO_3 and evaporated to give an oil. After flash chromatography on silica gel, eluted with a mixed solvent of C_2H_5OAc and methanol, 0.61 g of C VIII is obtained (50% yield).

^{1}H-NMR ($CDCl_3$, 300 MHz) 7.18 (1H, brt, J=8.1), 6.81 (1H, dd, J_1=2.3, J_2=8.3), 6.77 (2H, m), 6.73 (1H, m), 6.35 (1H, d, 5=8.3), 4.79 (1H, brs), 4.68 (1H, brs), 4.11 (1H, s), 3.84 (3H, s), 3.81 (1H, m), 3.77 (1H, m), 2.92 (3H, s), 2.84 (2H, m), 2.70 (1H, m), 2.65 (1H, m), 2.54 (3H, s), 2.05 (2H, m), 1.42 (3H, s).

Selected ^{13}C-NMR

149.58, 143.29, 137.47, 126.81, 120.52, 116.23, 107.65, 116.23, 107.65, 106.59, 98.02, 55.33, 53.17, 52.59, 45.96, 60.85, 38.33, 37.12, 27.30.

MS (m/e, %): 408 (m^++ 1, 14), 407 (m^+, 57), 363 (15), 349 (4), 218 (14), 217 (100), 190 (34), 175 (6), 160 (55), 147 (6), 132 (21), 117 (7), 104 (5), 91 (9).

To a solution of HCl salt of C VIII (0.44 g, 1 mmol) in CH_2Cl_2 (10 ml) BBr_3 is added at 0°C. The solution is stirred at room temperature for 16 hours. 1 g of Na_2CO_3 and CH_3OH (5ml) are added to the mixture at 0-5°C. After 2 hours the solvent is evaporated. CH_2Cl_2 (60 ml) and water (20 ml) are added to the residue. The CH_2Cl_2 layer is collected and the

water layer is extracted with CH_2Cl_2. The combined organic phase is washed with brine, dried over Na_2CO_3. After evaporation of solvent, 0.32 g of product is obtained (81% yield).

^{1}H-NMR ($CDCl_3$, 300 MHz) 7.03 (1H, br.m), 6.82 (1H, dd, J_1=2.4, J_2=8.3), 6.77 (1H, d, J=2.4), 6.67 (1H, d, J=8.7), 6.56 (1H, br.d, J=8.0), 6.35 (1H, d, J=8.3), 4.78 (1H, brs), 4.67 (1H, brs), 4.15 (1H, s), 3.85 (1H, m), 3.78 (1H, m), 2.92 (3H, s), 2.83 (2H, m), 2.73 (1H, m), 2.63 (1H, m), 2.54 (3H, s), 1.95 (2H, dd, J_1=5.2, J_2=7.4), 1.42 (3H, s).

Selected ^{13}C-NMR ($CDCl_3$, 75 MHz):
154.84, 154.11, 149.55, 143.36, 137.38, 134.36, 126.76, 120.60, 117.95, 116.?5, 112.68, 106.71, 97.70, 77.19, 53.05, 52.68, 46.03, 41.89, 40.65, 38.03, 37.29, 27.24, 22.87.

Example 5

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dimethoxy-2(1*H*)-isoquinolinecarboxylate (C VI).

To a solution of eseroline (0.75 mmol) in CH_2Cl_2 (0.8 ml) was added 1,1'-carbonyldiimidazole (0.91 mmol). The mixture was stirred at room temperature for 45 min forming (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl imidazolecarboxylate after which 6,7-dimethoxy-1,2,3,4-tetrahydroisoquinoline (1.10 mmol) in CH_2Cl_2 (1 ml) was added. After 15 hours the reaction mixture was concentrated to give a residue, which was purified by column chromatography (C_2H_5OAc:CH_3OH:$(C_2H_5)_3N$,4:1:0.05) giving (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dimethoxy-2(1*H*)-isoquinolinecarboxylate (C VI)(254 mg, 0.58 mmol, 77%) as a white foam.

Example 6

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dihydroxy-2(1*H*)-isoquinolinecarboxylate (C VII).

The hydrochloride salt of (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dimethoxy-2(1*H*)-isoquinolinecarboxylate (C VI) (0.58 mmol) was suspended in CH_2Cl_2 (2 ml) and cooled to 0° C. Boron tribromide (727 mg, 2.90 mmol) was added dropwise and stirred for 30 min at 0° C and then 20 hours at room temperature. The mixture was cooled and triethylamine (0.5 ml) and methanol (0.5 ml) was added and stirred for 30 min at 10° C, concentrated and the residue was extracted between CH_2Cl_2:$NaHCO_3$, the organic phase was washed with NaCl(aq). Dried ($NaSO_4$) and concentrated to give a red syrup (173 mg). The residue was purified on a silica gel column (C_2H_5OAc:CH_3OH:$(C_2H_5)_3N$,4:1:0.05) giving (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dihydroxy-2(1*H*)-isoquinolinecarboxylate (CVII)(110 mg, 46%).

^{13}C-NMR ($CDCl_3$) d 155.1, 149.3, 144.0, 143.9, 143.7, 137.4, 125.5, 125.4, 124.1, 123.8, 120.7, 116.3, 115.0, 114.8, 112.7, 106.7, 97.7, 53.0, 52.6, 45.6, 45.4, 42.3, 41.8, 40.2, 38.2, 36.8, 28.2, 27.8, 27,0

MS 409 (28, M^+), 218 (79), 217 (100), 161 (66), 160 (86)

Biological Experiment 1

Determination of the inhibition of electric eel AChE activity.

Principle: AChE catalyses the hydrolysis of acetylthiocholine. The product, thiocholine, reacts with 4,4'-dithiopyridine to form 4-thiopyridone. The rate of formation of 4-thiopyridone is followed spectrophotometrically at 324 nm. The registration is continuous, and the rate of formation of 4-thiopyridone is recorded.

Apparatus, chemicals and reagents: AChE from electric eel. A stock solution of 20 U/ml is prepared in phosphate buffer, pH 7.4, and portioned into 3 ml vials and kept at -18 °C until

the time of analysis. On each day of analysis, a new sample is slowly thawed on ice and kept on ice until time for incubation.

Solutions of the test and reference substances were stock solutions of 20 ±1 µM in sodium phosphate buffer pH 2.5.

Incubation procedure for determining the 50% inhibitory concentration (IC_{50}): All activities are calculated as % of zero time value and with correction made for the decrease in activity for AChE in blanks during the incubation. In a 3 ml glass tube: 0.05 M sodium phosphate buffer pH 7.4, AChE (electric eel) 1 U/ml and test substance in concentrations ranging from 0.26 nM to 121 µM in a final volume of 2 ml. The initial AChE activity is determined and the tube is placed in a water bath at 37°C. After 180 min the AChE activity is measured again. Results are expressed as IC_{50} values, the concentration giving 50% inhibition of the enzyme activity.

Spectrophotometrical analysis: All measurements were carried out in 1.5 ml plastic cuvettes. To 850 µl PDS buffer (0.1 mM 4,4'-dithiopyridine, 37°C), 50 µl of the incubate is added. Recording begins on adding 100µl 10 mM acetylthiocholine (ASCh). The absorbance is followed at 324 nm for 2 min and the results are calculated from the difference in absorbance per minute (ΔA/min) values.

Measurement of spontaneous hydrolysis: A cuvette filled with 900 µl of 37°C 4,4'-dithiopyridine (PDS) is placed in the spectrophotometer. 100 µl of 10 mM ASCh is added and recording begins.

The compounds may also be tested <u>in vitro</u> for AChE inhibitory activity in brain homogenate from rat according to the following experiment.

Biological experiment 2

Determination of inhibition of rat brain AChE

The materials and method were the same as described in Biological experiment 1 except that in the 3 ml glass tube 20 µl of stock solution (20 ±1 µM) of substance is added to brain homogenate giving a final volume concentration of 200 nM of the substance. Results

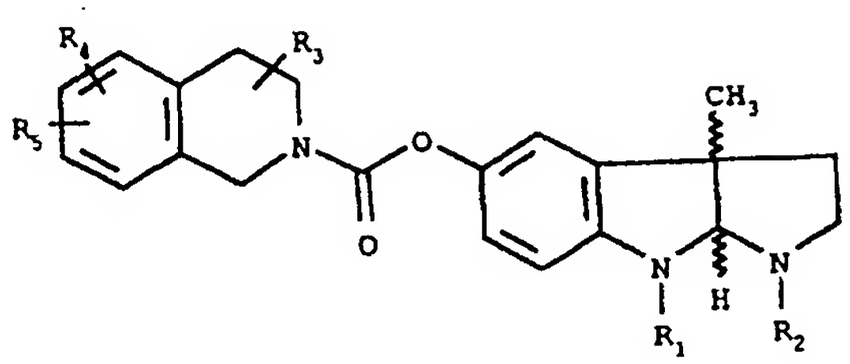
were recorded as % of control enzyme activity at 30, 60, 120 and 180 minutes, the control being incubation of rat brain homogenate without test substance. Thus, a low figure of AChE activity means that the substance is effective as an inhibitor of AChE.

Sample preparation of brain homogenate: Brain from Sprague-Dawley rat is weighed and homogenised in 19 parts of phosphate buffer 0.05 M, pH 8.0 containing 0.1% Triton X-100 (t-octyl phenoxypolyethoxyethanol) by use of a Heidolph Elektro homogenizer. The homogenate is transferred to plastic vials and kept on ice until analysed (within 10 min.). After analysis the vial is kept at -80°C .

The title compounds of Examples 1 to 4 were tested according to Biological Experiments 1 and 2 and were all found to be active.

Claims

1. The 3a*S-cis* and 3a*R-cis* isomers of a compound of the formula



I

wherein R_1 and R_2 , which may be the same or different are each is H or CH_3, and

R_3 is H or OH,

R_4 and R_5 , which may be the same or different, are each H, OH or OCH_3,

provided that

a) at least one of R_1 and R_2 is H when R_3 , R_4 and R_5 all are H,

b) when R_1 and R_2 are both CH_3 and R_3 is OH, then R_4 and R_5 are both H,

c) when R_1 and R_2 are both CH_3 and R_3 and R_4 are both H, then R_5 is OH in position 5 of the isoquinolyl moiety of the molecule,

d) when R_1 and R_2 are both CH_3 , and R_3 is H, then R_4 and R_5 are either both OH or both OCH_3 in positions 6 and 7 of the isoquinloyl moiety of the molecule,

racemic mixtures and any other mixture thereof, and pharmaceutically acceptable acid addition salts thereof.

2. Compounds according to claim 1, which are 3a*S* isomers.

3. Compounds according to claim 1, which are 3a*R* isomers.

4. The compound according to any of claims 1 or 2, which is

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate,

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolincarboxylate,

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-4-hydroxy-2(1*H*)-isoquinolinecarboxylate,

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-5-hydroxy-2(1*H*)-isoquinolinecarboxylate,

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dimethoxy-2(1*H*)-isoquinolinecarboxylate,

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dihydroxy-2(1*H*)-isoquinolinecarboxylate,

(3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-3a-methylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate.

5. A pharmaceutical composition comprising as active ingredient a compound as defined in any of claims 1 to 4.

6. A pharmaceutical composition for oral administration comprising as active ingredient a compound as defined in any of claims 1 to 4.

7. A pharmaceutical composition for parenteral administration comprising as active ingredient a compound as defined in any of claims 1 to 4.

8. A compound according to any of claims 1 to 4 for use in therapy.

9. A compound according to any of claims 1 to 4 for use in the treatment of diseases characterized in a cholinergic deficit.

10. A compound according to any of claims 1 to 4 for use in the treatment of diseases characterized in memory dysfunction.

11. A compound according to any of claims 1 to 4 for use in the treatment of Alzheimer's disease.

12. Use of a compound according to any of claims 1 to 4 for the preparation of a medicament for alleviating a cholinergic deficit.

13. Use of a compound according to any of claims 1 to 4 for the preparation of a medicament for alleviating memory dysfunction.

14. Use of a compound according to any of claims 1 to 4 for the preparation of a medicament for alleviating Alzheimer's disease.

15. A method of treating a patient in need of alleviation of a cholinergic deficit, which method comprises administering to such a patient an effective amount of a compound as defined in any of claims 1 to 4.

16. A method of treating a patient in need of alleviation of memory dysfunction, which method comprises administering to such a patient an effective amount of a compound as defined in any of claims 1 to 4.

17. A method of treating a patient in need of alleviation of Alzheimer's disease, which method comprises administering to such a patient an effective amount of a compound as defined in any of claims 1 to 4.

18. A process for manufacturing a compound according to claim 1, wherein R_1, R_3, R_4, and R_5 all are H and R_2 is CH_3, characterized in treating (3a-*cis*)-8-formyl-1,2,3,3a,8,8a-hexahydro-1,3a-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)isoquinolinecarboxylate with a strong acid in aqueous solution.

19. A process for manufacturing a compound according to claim 1, wherein R_2, R_3, R_4, and R_5 all are H, and R_1 is CH_3, characterized in treating (3a-*cis*)-1,2,3,3a,8,8a-hexahydro-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate with hydrogen in the presence of a palladium catalyst.

20. A process for manufacturing a compound according to claim 1 wherein R_1 and R_2 are both CH_3 , R4 and R_5 are H, and R_3 is OH in position 1, 3 or 4 of the isoquinoline moiety of the molecule, characterized in reacting eseroline with carbonyldiimidazole and 1,2,3,4-tetrahydro-hydroxyisoquinoline.

21. A process for manufacturing a compound according to claim 1, wherein R_1 and R_2 both are CH_3, R_3 is H, and one of R_4 or R_5 is OH in any of positions 5, 6, 7, or 8 of the isoquinoline moiety of the molecule the other substituent of R_4 or R_5 being H , characterized in demethylating (3a-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-methoxy-2(1*H*)-isoquinolinecarboxylate

having the methoxyl group in any of positions 5, 6, 7, or 8 if the isoquinoline moiety of the molecule with a mineral acid or a Lewis acid.

22. A process for manufacturing a compound according to claim 1, wherein R_1 and R_2 both are CH_3, R_3 is H, and R_4 and R_5 are both methoxy characterized in treating eseroline with carbonyldiimidazole.

23. A process for manufacturing a compound according to claim 1, wherein R_1 and R_2 both are CH_3, R_3 is H, and R_4 and R_5 are both hydroxy characterized in treating the hydrochloride salt of (3a*S*-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-6,7-dimethoxy-2(1*H*)-isoquinolinecarboxylate with boron tribromide and triethylamine.

24. (3a-*cis*)-1-formyl-1,2,3,3a,8,8a-hexahydro-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1*H*)-isoquinolinecarboxylate.

25. (3a-*cis*)-1,2,3,3a,8,8a-hexahydro-1-benzyl-3a,8-dimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-2(1 *H*)isoquinolinecarboxylate.

26. (3a-*cis*)-1,2,3,3a,8,8a-hexahydro-1,3a,8-trimethylpyrrolo[2,3-*b*]indol-5-yl 3,4-dihydro-methoxy-2(1*H*)-isoquinolinecarboxylate having the methoxyl substituent in any of positions 5, 6, 7, or 8 of the isoquinolyl moiety of the molecule.

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 96/01679

A. CLASSIFICATION OF SUBJECT MATTER

IPC6: C07D 487/04, A61K 31/40
According to International Patent Classification (IPC) or to both national classification and IPC

B. FIELDS SEARCHED

Minimum documentation searched (classification system followed by classification symbols)

IPC6: C07D

Documentation searched other than minimum documentation to the extent that such documents are included in the fields searched

SE,DK,FI,NO classes as above

Electronic data base consulted during the international search (name of data base and, where practicable, search terms used)

CAS-ONLINE

C. DOCUMENTS CONSIDERED TO BE RELEVANT

Category*	Citation of document, with indication, where appropriate, of the relevant passages	Relevant to claim No.
A	US 5187165 A (RUSSELL R.L. HAMER ET AL), 16 February 1993 (16.02.93) --	1-17,21-29

☐ Further documents are listed in the continuation of Box C. ☒ See patent family annex.

* Special categories of cited documents:
"A" document defining the general state of the art which is not considered to be of particular relevance
"E" erlier document but published on or after the international filing date
"L" document which may throw doubts on priority claim(s) or which is cited to establish the publication date of another citation or other special reason (as specified)
"O" document referring to an oral disclosure, use, exhibition or other means
"P" document published prior to the international filing date but later than the priority date claimed

"T" later document published after the international filing date or priority date and not in conflict with the application but cited to understand the principle or theory underlying the invention
"X" document of particular relevance: the claimed invention cannot be considered novel or cannot be considered to involve an inventive step when the document is taken alone
"Y" document of particular relevance: the claimed invention cannot be considered to involve an inventive step when the document is combined with one or more other such documents, such combination being obvious to a person skilled in the art
"&" document member of the same patent family

Date of the actual completion of the international search	Date of mailing of the international search report
30 January 1997	01 -03- 1997
Name and mailing address of the ISA/ Swedish Patent Office Box 5055, S-102 42 STOCKHOLM Facsimile No. +46 8 666 02 86	Authorized officer Göran Karlsson Telephone No. +46 8 782 25 00

Form PCT/ISA/210 (second sheet) (July 1992)

INTERNATIONAL SEARCH REPORT

International application No.
PCT/SE 96/01679

Box I Observations where certain claims were found unsearchable (Continuation of item 1 of first sheet)

This international search report has not been established in respect of certain claims under Article 17(2)(a) for the following reasons:

1. [X] Claims Nos.: 18-20
because they relate to subject matter not required to be searched by this Authority, namely:
A method for treatment of the human or animal body by therapy, see rule 39.1.

2. [ ] Claims Nos.:
because they relate to parts of the international application that do not comply with the prescribed requirements to such an extent that no meaningful international search can be carried out, specifically:

3. [ ] Claims Nos.:
because they are dependent claims and are not drafted in accordance with the second and third sentences of Rule 6.4(a).

Box II Observations where unity of invention is lacking (Continuation of item 2 of first sheet)

This International Searching Authority found multiple inventions in this international application, as follows:

1. [ ] As all required additional search fees were timely paid by the applicant, this international search report covers all searchable claims.

2. [ ] As all searchable claims could be searched without effort justifying an additional fee, this Authority did not invite payment of any additional fee.

3. [ ] As only some of the required additional search fees were timely paid by the applicant, this international search report covers only those claims for which fees were paid, specifically claims Nos.:

4. [ ] No required additional search fees were timely paid by the applicant. Consequently, this international search report is restricted to the invention first mentioned in the claims; it is covered by claims Nos.:

Remark on Protest

[ ] The additional search fees were accompanied by the applicant's protest.

[ ] No protest accompanied the payment of additional search fees.

INTERNATIONAL SEARCH REPORT

Information on patent family members

28/10/96

International application No.
PCT/SE 96/01679

Patent document cited in search report	Publication date	Patent family member(s)		Publication date
US-A- 5187165	16/02/93	AT-T-	112278	15/10/94
		AT-A-	260287	15/04/95
		AT-B-	400331	27/12/95
		AU-B-	612583	18/07/91
		AU-A-	7566887	21/01/88
		CA-A-	1307788	22/09/92
		CA-A-	2085216	28/12/91
		DE-D,T-	3750596	23/03/95
		EP-A,B-	0253372	20/01/88
		SE-T3-	0253372	
		ES-T-	2060585	01/12/94
		IE-B-	64429	09/08/95
		KR-B-	9603614	20/03/96
		KR-B-	9606064	08/05/96
		US-A-	4791107	13/12/88
		US-A-	5541216	30/07/96
		US-A-	5541340	30/07/96
		US-A-	5547977	20/08/96
		US-A-	5550253	27/08/96
		US-A-	5550254	27/08/96